Remarks

The Office Action mailed April 14, 2008, has been received and reviewed. Claim 83 is amended to correct a clerical error. The pending claims are claims 75, 76, 78, 79, and 82-98. Reconsideration and withdrawal of the rejections are respectfully requested.

The 35 U.S.C. §112, First Paragraph, Rejection

The Examiner has rejected claims 75, 76, 78, 79, and 82-98, under 35 U.S.C. §112, first paragraph, alleging the specification does not enable any person skilled in the art to which it pertains, or with which it is most nearly connected, to use the invention commensurate in scope with these claims. This rejection is respectfully traversed.

The standard for determining whether the specification meets the enablement requirement was cast in the Supreme Court decision of Mineral Separation v. Hyde, 242 U.S. 261, 270 (1916) which postured the question: is the experimentation needed to practice the invention undue or unreasonable? That standard is still the one to be applied. In re Wands, 858 F.2d 731, 737, 8 USPQ2d 1400, 1404 (Fed. Cir. 1988).

The Examiner sets forth three bases for lack of enablement, asserting the specification (i) does not reasonably provide enablement for use of double stranded RNAs (dsRNAs) targeted to any other foreign, endogenous or pathogen genes in all types of vertebrate cells, (ii) does not reasonably provide enablement for an *ex vivo* method that includes treatment of an explanted cell with dsRNA followed by implantation into an organism, and (iii) does not reasonably provide enablement for using the *ex vivo* method to treat a disease or pathogen. Office Action at page 3.

I. Enablement for use of dsRNAs targeted to any other foreign, endogenous or pathogen genes in all types of vertebrate cells.

To support the assertion that the claims are not enabled for use of dsRNAs in all types of vertebrate cells, the Examiner argues that the results observed in the working examples have limited applicability to the broad scope of the claims. Specifically, "[b]ecause the cells used in

the working examples lack a full immune response the working examples cannot be extrapolated to cells having the PKR response." Office Action at page 7.

The Examiner is requested to note that the non-specific response to dsRNAs is not dependent on having an immune system, but rather it is dependent on whether the particular cell expresses the necessary components of the pathway, such as the PKR kinase and RNase L. This response is a cellular response, not a systemic response.

The skilled person knew that a full immune response was not required for individual cells to mount a PKR response. Inhibition of protein synthesis is one of the characteristics of a PKR response, and this was known to occur in cell extracts. For example, Tuschl et al. (1999, Genes and Development, 13:3191-3197, of record) report the addition of dsRNA to rabbit reticulocyte lysate caused a profound and rapid, nonspecific decrease in mRNA stability. Tuschl et al., page 3195, left-hand column. Inhibition of protein synthesis was also known to occur in intact cell cultures. For example, Cordell-Stewart and Taylor (1973, Journal of Virology, 11:232-237, copy included), report the addition of dsRNA to intact mouse Ehrlich ascites tumor cells in culture results in a rapid cessation of cellular polypeptide synthesis. Cordell-Stewart and Taylor, page 232, left-hand column. These documents show that the skilled person knew a full immune response was not necessary for a cell to exhibit a PKR response. Thus, the Examiner's conclusion that "[b]ecause the cells used in the working examples lack a full immune response the working examples cannot be extrapolated to cells having the PKR response" is unfounded.

Further, the skilled person knew that zebrafish embryos would mount a PKR response. Microinjection of RNA in zebrafish embryos has been shown to produce several unspecific defects. Hyatt and Ekker (1999, In Methods in Cell Biology (Detrich et al., eds.), pages 117-126, Academic Press, San Diego, CA, copy included). The presence of a PKR response in zebrafish embryos has been documented by others. Oates et al. (2000, Developmental Biology 224: 20-28, of record) reported that dsRNA injected into early zebrafish embryos produced a nonspecific depletion of several endogenous mRNAs and concluded that "RNAi appears unsuited to application in the zebrafish embryo...." (page 21, left-hand column). Likewise, Zhao et al. (2001,

Developmental Biol., 229:215-225, of record) also report that injection of dsRNA results in degradation of endogenous mRNA and has a nonspecific effect at the posttranscriptional level. The non-specific depletion of endogenous mRNA in response to exogenous dsRNA is a hallmark of a PKR response. The results of Oates et al. and Zhao et al. show that zebrafish embryos have the ability to mount a PKR response when dsRNA is injected into a zebrafish embryo. Oates et al. and Zhao et al. are post-filing date documents; however, in view of Hyatt and Ekker, these documents provide evidence of what one skilled in the art would have known on or before the effective filing date of the patent application. M.P.E.P §2164.05(a). Thus, the skilled person would have known that zebrafish embryo cells could mount a PKR response. Again, the Examiner's conclusion that "[b]ecause the cells used in the working examples lack a full immune response the working examples cannot be extrapolated to cells having the PKR response" is unfounded.

The Examiner notes that thymus, an organ involved in the immune system, is derived from neural crest tissue, and based on this premise concludes that the skilled person would expect that this tissue is also unable to mount an immune response to dsRNA. As discussed above, the skilled person knew on or before the effective filing date of the present application that individual cells and even cell extracts will mount a response to dsRNA. The skilled person would not draw the conclusion made by the Examiner, but would conclude that neural crest tissue is likely to act like other cells and mount a PKR response when dsRNA is introduced.

The Examiner also asserts that NIH3T3 cells do not exhibit the PKR response in the same way as adult cells. NIH3T3 cells were not used in the working examples. The specification states that the experiments in Example 3 were performed in mouse NIH/3T3 cells. However, in the preliminary amendment dated April 28, 2003, a declaration by Dr. Yin-Xiong Li indicates that the cells used for the experiments in Example 3 were actually rat ROS cells and were inadvertently labeled as mouse NIH/3T3 cells (see paragraph 4 of the declaration dated April 22, 2003). Moreover, Caplan et al. (2002, Gene, 252: 95-105, of record) describes experiments with three different mammalian cell culture lines, including NIH3T3 cells, showing that no sequence

specific knockdown of mRNA was observed in these cells, which they attribute to the PKR response. Caplen et al., page 103, first full paragraph. Likewise, Elabashir et al. (2001, Nature, 411:494-498, of record) demonstrate that introduction of long dsRNAs into NIH3T3 cells non-specifically reduced reporter-gene expression. Elabashir et al., page 496, first column, second full paragraph.

The Examiner is also requested to consider Carter and DeClercq (1974, Science, 186: 1172-1178, of record) for further evidence that individual cells, tissues, and embryos mount a PKR response to dsRNA. Carter and DeClercq disclose that dsRNA induces interferon activity in chick embryo, chick liver, rat liver, rabbit kidney, and HELA cells. Carter and DeClercq, page 1173, first column, point 2.

Wianny et al. (2000, Nature Cell Biology, 2:70-75, of record) is cited by the Examiner to further support the assertion that the results observed in the examples are not broadly applicable to all vertebrate cells. Wianny et al. state that "it is possible that the early mouse embryo is incapable of an interferon response and that there may still be difficulties in using RNAi at later stages" (p. 73, under Discussion). The caveats that "it is possible" and "there may still be difficulties" indicate that this is hardly definitive language regarding whether the results observed in the examples are broadly applicable to all vertebrate cells, especially in view of other post-filing date documents such as Oates et al. and Zhao et al.

The applicants note that the claims do not recite language requiring the absence of a PKR response. The issues raised by the Examiner regarding the need for the specification to teach how to avoid the PKR response are not pertinent, and do not render the claimed invention unpredictable. Thus, any perceived unpredictability based on induction of a PKR response or post-filing date evidence that short dsRNA may be preferred in some aspects is irrelevant with respect to evaluating the enablement of the claimed invention.

II. Enablement for an *ex vivo* method that includes treatment of an explanted cell with dsRNA followed by implantation into an organism.

The Examiner cites several documents to support the assertion that histoincompatible allografts and xenografts are rejected by the host. Rejection of histoincompatible tissue by a host is well known, and the antigens on cells that contribute to histoincompatibility, such as HLA antigens, are known. The skilled person can easily evaluate known antigens present on a donor cell and the likelihood such antigens will result in rejection in the recipient. Likewise, methods for minimizing rejection are known and include, for instance, immunosuppression and encapsulation. Gage, 1998, Nature, 392Suppl:18-24, Table 1 (of record). The ability to evaluate histocompatibility and control rejection of histoincompatible tissue is apparent in view of the prevalent use of allografts. Allografts are routine, and include, for instance, tissues such as skin, cornea, heart, liver, kidney, bone, and ligament, and cells such as bone marrow cells, islets of Langerhans, and blood cells. Xenografts are more limited, but also routine, and include porcine heart valve transplants into humans. One document cited by the Examiner, Gage (1998, Nature, 392Suppl:18-24), discloses solutions to allogeneic and xenogeneic tranplantations, such as immunosuppression and encapsulation. Gage, Table 1.

The Examiner asserts that methods for reducing host rejection of an allogeneic or xenogeneic cell transplant would need to be empirically determined. To support this assertion, the Examiner cites Platt (1998, Nature, 392Suppl:11-17) as teaching that a skilled artisan would need to know how to prevent infection of the host organism while the host's immune system is suppressed. Platt et al. actually state that "[a]dvances in clinical care have vastly improved the outcome of organ transplantation." Platt at page 14, in box 1. Regarding the Examiner's assertion that the specification does not teach how to use immunosuppressive drugs in the context of the present claims, the use of immunosuppressive drugs following transplantation is routine, and an applicant need not disclose in detail, and preferably omits, that which is conventional or well known in the art. M.P.E.P. §2163(II)(A)(2).

The claims may encompass allogeneic and xenogeneic transplantation, but it is the applicants' position that the skilled person can determine if an allogeneic or xenogeneic transplantation will be successful using routine experimentation.

Claim 76 is directed to syngeneic transplantation, i.e., cells that are deviod of any histoincompatibility. According to the Examiner, the use of syngeneic cells is not enabled because a skilled artisan would need to be taught what kind of cells should be isolated, how to isolate the cells, and how to culture the cells. The applicant respectfully disagrees that the specification must teach each of these. Determining what cell should be isolated, and how to isolate and culture such cells is routine in the art of transplantation. The fact that experimentation may be complex does not necessarily make it undue, if the art typically engages in such experimentation. M.P.E.P. §2164.01. Further, methods for syngeneic transplantation have been routine in the art for at least 40 years. For instance, bone marrow cells are routinely removed from and infused back into patients during treatments that destroy dividing cells, such as chemotherapy. Thomas et al., 1957, N. Engl. J. Med., 157:491-496, and Philip et al. 1995, N. Engl. J. Med., 333:1540-1545, copies included.

Further, regarding possible rejection of a transplanted cell, the applicants note that these types of issues have been raised under the "how to use" aspect of 35 U.S.C. §112 for pharmaceutical use in humans. The issue was addressed in *In re Krummel*, 292 F.2d 948, 130 U.S.P.Q. 215 (C.C.P.A 1961), where the court stated:

> "There is nothing in the patent statute ... which gives the Patent Office the right or the duty to require an applicant to prove that compounds or other materials which he is claiming ... are safe, effective, and reliable for use with humans."

This issue was further addressed in *In re Hartop*, 311 F.2d 249, 135 U.S.P.Q. 419 (C.C.P.A. 1962) in which that CCPA held that absolute safety was not required, stating:

> "Congress has recognized this problem and has clearly expressed its intent to give statutory authority and responsibility in this area to Federal agencies different than that given to the patent office."

As this is the standard applied for pharmaceutical use in humans, which one would expect to be the highest standard possible, the applicants in the present case should not be required to prove the absence of possible immune responses when claims are directed to attenuation of gene

expression in vertebrate cells. This is particularly true in light of the lack of terms regarding toxicity or immunogenicity in applicants' claims.

The present invention includes explanting a vertebrate cell from a vertebrate organism, supplying the cell with at least one double stranded RNA, and implanting the cell into a vertebrate organism, wherein expression of the target gene is attenuated in the vertebrate cell. The use of allogeneic and xenogeneic cells in transplants may require experimentation, but the test of enablement is not whether any experimentation is necessary, but whether, if experimentation is necessary, it is undue. M.P.E.P. §2164.01. The testing required to use allogeneic or xenogeneic cells in a transplantation may require evaluating the histocompatibility of the donor and recipient, but this and other steps in an allogeneic or xenogeneic transplantation are routine. Moreover, complications with host rejection in allogeneic and xenogeneic transplants can be controlled using methods known in the art, and complications with host rejection in syngeneic transplants do not exist.

III. Enablement for using the *ex vivo* method to treat a disease or pathogen.

The Examiner asserts that claims 96-98 are directed to treatment of a disease by transplanting cells. The claims are not drawn to treating a disease or pathogen. Rather, the claims are directed to a method of attenuating the expression of a target gene in a vertebrate cell *ex vivo*, wherein the target gene can be a gene associated with a disease or pathogen. Thus, the Examiner's reliance on Coburn et al. (J. Antimicrob. Chemother., 2003, 51:753-756, of record) is irrelevant. A skilled artisan using the guidance in the specification could design an appropriate dsRNA to target a gene associated with a disease or pathogen and use it to attenuate expression of that gene in a vertebrate cell *ex vivo*.

Summary

The applicants note that the presumption is that an application is enabled, and that this is overcome only if the Examiner can show that undue experimentation is necessary to use the invention as claimed. Furthermore, the mere fact that experimentation may be involved, and even be complex, does not necessarily make the experimentation undue. Undue experimentation would not be necessary to practice the invention as claimed. It is well-established that some experimentation is often to be expected in unpredictable technologies, such as molecular biology. The question is whether the amount of experimentation needed to practice the invention, as claimed, is undue. This question is answered more readily if the method of the invention is broken down into separate steps. For the first step, a vertebrate cell is explanted from a vertebrate organism. Methods for accomplishing this are routine and known to those skilled in the art. Next, to attenuate the expression of a target gene in a vertebrate cell, the nucleotide sequence of the gene can be obtained either from a database or from routine procedures used to determine the sequence of a gene. RNA capable of hybridizing to and silencing the target gene is then synthesized, again using routine methods known to those skilled in the art as well as the direction and guidance provided by the present specification. The dsRNA is supplied to a vertebrate cell using methods which are again routine and known to those skilled in the art. Finally, the cell is implanted into a vertebrate organism, using routine methods known to those skilled in the art. It is thus respectfully submitted that the pending claims are enabled. Reconsideration and withdrawal of the rejection of the pending claims under 35 U.S.C. §112, first paragraph, is accordingly requested.

The 35 U.S.C. §102 Rejection

The Examiner has rejected claims 75, 76, 78, 79, 82-91 and 93-98, under 35 U.S.C. §102(e) as being anticipated by Fire et al. (U.S. Patent 6,506,559). This rejection is respectfully traversed.

To begin with, the Examiner asserts that the level of enablement of the present specification and the level of enablement of Fire et al. are equivalent. Office Action, page 20, first paragraph. In the rejection for lack of enablement detailed in the Office Action, the Examiner asserts that the present specification is enabling only for what is disclosed in the working examples. Thus, it appears that the Examiner considers both the present specification and Fire et al. to enable only what they disclose in working examples. In other words, it is the Examiner's view that the present specification enables the use of the vertebrate cells as disclosed in the working examples and nothing more, and that Fire et al. enable the use of *C. elegans* as disclosed in the working examples and nothing more. The Examiner appears to support the non-enablement of Fire et al. as it relates to explanting vertebrate cells, treating the explanted vertebrate cells with dsRNA, and implanting the cells into a vertebrate organism.

The applicants presented detailed arguments in the Appeal Brief relating to the present rejection under 35 U.S.C. §102(e). The dismissal of those arguments is based on the Examiner's belief that the claims are not enabled. Specifically, the Examiner notes that "the examples described in the instant specification were performed in embryos and embryonic tissues that do not have an immune response or have an immune response likely to be compromised, therefore they do not provide a disclosure commensurate in scope with the instant claims." Office Action, page 19. As discussed in detail in the immediately preceding section, the Examiner is not correct; the working examples use cells and tissues that can mount a PKR response to dsRNA. Thus, the Examiner cannot use this premise as the basis for the rejection under 35 U.S.C. §102(e). Since this premise is false and cannot be used as the basis for the rejection under 35 U.S.C. §102(e), the present rejection should be withdrawn.

The Examiner is respectfully requested to reconsider the comments made in the Appeal Brief and withdraw the rejection of the pending claims as anticipated by Fire et al. (U.S. Patent 6,506,559).

The 35 U.S.C. §103 Rejection

The Examiner has rejected claims 75, 76, 78, 79, and 82-98 under 35 U.S.C. §103(a) as being unpatentable over Fire et al., in view of Ekenberg et al. (Promega Notes Magazine Number 46, 1994, pages 14-17). This rejection is respectfully traversed.

The Examiner notes that claims 75, 76, 78, 79, 82- 91, and 93-98 have been rejected as obvious because they have been rejected under §102(e) as anticipated by Fire et al. Office Action, page 24, first full paragraph. "Rejections on obviousness cannot be sustained by mere conclusory statements; instead, there must be some articulated reasoning with some rational underpinning to support the legal conclusion of obviousness." M.P.E.P. §2141(III). The assertion that the pending claims are obvious because they are anticipated is conclusory, and does not support the rejection of the pending claims as obvious in view of the cited art. The Examiner does not set forth a *prima facie* case for the obviousness of claims 75, 76, 78, 79, 82- 91, and 93-98 in view of the cited art.

The applicants maintain that one of ordinary skill in the art would not be motivated to modify the methods disclosed in Fire et al. with those disclosed in Ekenberg et al. or any other cited reference for application in vertebrate cells with a reasonable expectation of success. The Examiner is respectfully requested to reconsider the comments made in the Appeal Brief and withdraw the rejection of the pending claims as obvious over Fire et al., in view of Ekenberg et al.

Summary

It is respectfully submitted that the pending claims are in condition for allowance and notification to that effect is respectfully requested. The Examiner is invited to contact Applicants' Representatives at the telephone number listed below if it is believed that prosecution of this application may be assisted thereby.

Respectfully submitted
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CERTIFICATE UNDER 37 CFR §1.8:
The undersigned hereby certifies that the paper(s), as described hereinabove, are being transmitted via the U.S. Patent and Trademark Office electronic filing system in accordance with37 CFR §1.6(a)(4) to the Patent and Trademark Office addressed to the Commissioner for Patents, Mail Stop, P.O. Box 1450, Alexandria, VA 22313-1450, on this 14 day of October, 2008.

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